Induced breeding of farm-bred and pond-raised critically endangered peninsular carp, *Hypselobarbus pulchellus*

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Adult Hypselobarbus pulchellus.

Puntius pulchellus also called *Hypselobarbus pulchellus* is endemic to the peninsular rivers of India, mainly the Krishna, Godavari, Tungabhadra, Sita and Tunga. It once formed a major fishery of the Tungabhadra reservoir but has declined to the status of a critically endangered fish species. *H. pulchellus* is a bentho-pelagic species which inhabits the deeper part of large streams and rivers along the base of ghats. Pulchellus is considered to be the only indigenous fish consuming aquatic weeds and submerged grasses and this fish could be used in controlling aquatic vegetation in reservoirs, tanks and irrigation canals. Though herbivorous, pulchellus is known to change its feeding habits depending on the availability of food. This fish, which is capable of attaining 8 kg could become a welcome addition to pond culture practices of India, especially for composite fish culture.

The need for diversification of farmed fish species in recent years has resulted in renewed interest in the breeding, propagation and culture of *H. pulchellus*. This has resulted in successful induced breeding of wild stock of this species by the ICAR-Central Institute of Freshwater Aquaculture, paving the way for its introduction to aquaculture systems of the country (Sridhar et al., 2014). In natural waters, the breeding of *H. pulchellusis* reported to commence soon after the monsoon months from September which continued until April with a peak in September and January. However, under pond culture conditions, we obtained first maturity of both the sexes during June-July and breeding of this fish continued till October of the same year. This article gives an overview of the breeding and seed production technologies developed by ICAR-CIFA for farm bred and pond raised *H. pulchelllus*.

Brood stock management

Brood stock was raised from the F1 generation of this fish produced at our research centre through induced breeding of pulchellus collected from the wild and reared under pond conditions. Broodstock ponds of 0.01 ha area were stocked with males and females weighing around 800-1200 g, three months prior to breeding season @ 2000 /ha (10 of each). The ponds were manured initially with cow dung @ 3-4t/ha, 7-10 days prior to stocking. This fish prefers clear waters and a minimum water depth of 1 meter was maintained. Pulchellus has a shoaling tendency and swims in groups and as such it is easy to observe the fish and record any abnormal behaviour/swimming pattern. The fish were fed diet with a formulated diet containing fishmeal (22%), groundnut oil cake (30%), rice bran (38%), finger millet (7%) and vitamin and mineral mixture 92%) having crude protein content of 35%. Feed was prepared as follows. All the ingredients were sieved through a fine meshed screen (0.5 mm) and mixed with hot water to make dough. The dough was cooled, vitamin-mineral mixture added and pressed through a pelletizer to get uniform

sized pellets (3 mm). The pellets were sun-dried and packed in air-tight plastic bags till further use.

Selection of brooders

The fish were observed regularly for maturity through periodic sampling. Farm bred fish attained first maturity at 1.5-2 years of age. It was easy to select brooders for induced breeding as sexual dimorphism was exhibited very distinctly by mature males and females. Males during breeding season were distinguished by a dark colour, especially at the abdominal region and a pink snout with prominent tubercles between the eyes spreading to the snout, in contrast to the white and swollen belly, plain and smooth snout and swollen pinkish vent of females. Male maturity was also judged by the production of milt, which got expressed on application of slight pressure on the lateral sides of the abdomen near the vent. The appearance of tubercles in males was seasonal and disappeared after 3–4 months of its onset. Out of the 10 females stocked, 7 attained maturity and were used for breeding.

Spawning

The fish was successfully bred using a preparation consisting of salmon gonadotropin releasing hormone analogue and domperidone (Ovatide). The first injection of the hormone preparation was given to both sexes at a dosage of 0.5 ml/kg body weight. All injections were given intramuscularly between the base of the dorsal fin and lateral line by lifting the fish scale to insert the needle. After injection and withdrawal of the needle the area was gently massaged to aid distribution of the hormone into the musculature and prevent any backflow. The injected brooders were released to the breeding pool of a Chinese hatchery and were allowed to remain there with circulating water and overhead shower running throughout. After 8–12 hours when the belly of the females became soft and swollen the second dose of hormone preparation was administered to both males and females at the same dose given earlier. After 12 hours of the second hormone administration, the fish were anaesthetized by submersing in 25 ppm solution of clove oil. The dry method of stripping was followed. First, the female fish were wiped gently with a dry and clean cloth and held in an inclined position with the head up and the ventral portion over the basin. A slight pressure was applied gently with the thumb and index finger on the swollen belly, slowly descending towards the lower end of the body down to the vent. The fully mature females released a stream of ripe eggs. Immediately after stripping the eggs, the procedure was repeated with the males and the milt directly stripped on to the eggs. The milt was allowed to fertilize the eggs by slow orbital rotation of the basin for a period of 10 min. The eggs were then washed with freshwater, till the washings become clear of the milt. The fertilized eggs appeared bright orange in colour. Since the fish is a batch spawner, the number of ova released per spawning was less (relative fecundity 8,029 + 3,539/ kg female) compared to other carps. Further, the fecundity recorded in the present study was lower compared to that reported by Sridhar et al. (2014) for the same species. The difference in the two studies was that Sridhar et al. (2014) used wild collected specimens for developing brooders, while in the present study, the brooders were developed from the fingerlings produced at the Centre. Kulkarni (2000) and Keshavanath et al. (2006) observed that in pond-raised mahseer the fecundity is comparatively low.

Above, male (note prominent turbercles across upper lip); below, female.

Incubation

After fertilisation, the bright orange eggs were transferred to a hatching unit specially designed for heavy yolk laden eggs. The unit consisted of a series of rectangular FRP tanks with plastic trays having a synthetic net bottom (1 mm mesh) with continuous flow of water on a re-circulatory mode with necessary bio filters to ensure good water quality. The fertilised eggs were spread uniformly on the trays and a water level of 2 inches was maintained over the eggs. The bad (white) eggs were removed daily. A fertilisation rate of 70.80% was recorded. The eggs attained a size of about 3mm in 48 hours. At this stage

Stripping female.

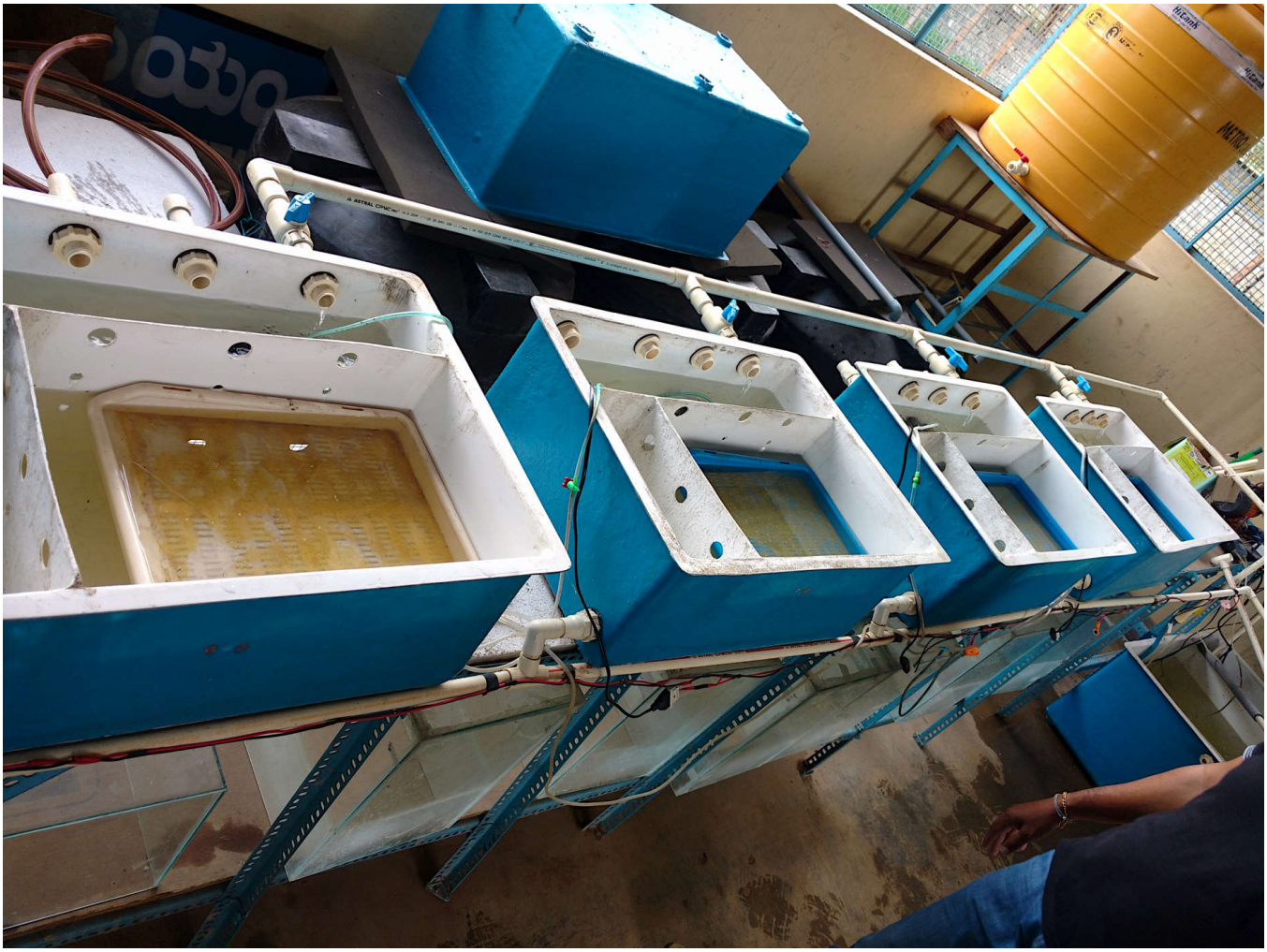
Stripping male.

Fertilised eggs.

Hatching.

Spawn.

Fry.



Recirculatory hatchery unit, note the hatching trays with eggs.

the eggs became translucent and the twitching larvae could be clearly seen inside when examined under an inverted microscope.

The development of the embryos was slow with the elongation of the yolk mass taking about 24 hours at water temperature of 22-24°C. As the embryo advanced in its development, the movements became more vigorous. The first larva hatched out after a period of 48 hours with a heavily laden yolk sac. A hatching rate of 68.06% was recorded. The yolk was completely absorbed at 5-6 days post hatching.

Rearing of hatchlings

After yolk absorption on day 6 post hatching the larvae were shifted to glass aquaria maintained at 26°C with constant aeration @ a stocking rate of 1,000/m3 of water volume and fed with filtered zooplankton for a period of 5 days followed by a combination of filtered zooplankton and finely ground pelleted feed containing 35% crude protein explained earlier, as additional supplementary feed for another 10 days. The dead larvae, if any, along with other uneaten feed and faecal matter at the tank bottom were removed daily before feeding. On every third day one third of the water from the tank was removed and replenished with fresh water. The fry thus obtained, 15 days post hatching, were transferred to nursery tanks for rearing to fingerlings.

Successful breeding of the farm bred and pond raised *H. pulchellus* will have implications on conservation of this endangered species as well as a providing a new candidate species for freshwater aquaculture.

References

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